463.34 M776n 29724 6/30/53 A.E.S.L.

NITROGEN NUTRITION OF TOMATOES WITH FOLIAR SPRAYS OF UREA

By

JAMES MONTELARO

A DISSERTATION PRESENTED TO THE GRADUATE COUNCIL OF
THE UNIVERSITY OF FLORIDA
IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE
DEGREE OF DOCTOR OF PHILOSOPHY

UNIVERSITY OF FLORIDA
August, 1952

ACKNOWLEDGMENTS

The author is greatly indebted to Dr. C. B. Hall, Assistant Horticulturist, Agricultural Experiment Station, under whose guidance this work was done. His help and advice were a constant source of encouragement throughout the course of this work.

To Dr. F. S. Jamison, Horticulturist, Agricultural Experiment Station, who suggested the problem and offered direction and assistance whenever it was needed, the author wishes to express his sincere thanks.

With deep gratitude he acknowledges the advice given to him in the pursuit of studies by Dr. H. S. Wolfe, Head of the Horticulture Department.

He also wishes to express his appreciation for the help received from Dr. G. J. Stout, Professor of Horticulture, Dr. T. W. Stearns, Associate Professor of Agricultural Chemistry, Dr. R. A. Dennison, Associate Horticulturist, Agricultural Experiment Station, and Dr. L. C. Hammond, Assistant Professor of Soils.

To the many other staff members of the University of Florida who gave unselfishly of their time, the author shall always be grateful.

29724

TABLE OF CONTENTS

		Page
I.	INTRODUCTION...............................	1
II.	REVIEW OF LITERATURE..........................	3
	A. Foliar Injury..........................	9
	B. Absorption of Urea.....................	10
	C. Assimilation of Urea...................	11
III.	EXPERIMENTAL..................................	13
	A. Response of Tomatoes to Urea Sprays.....	13
	1. Experiment I (Fall, 1950)........	14
	2. Experiment II (Spring, 1951).....	32
	3. Experiment III (Spring, 1952)....	41
	4. Experiment IV (Greenhouse, 1951).	44
	B. Studies on Foliar Injury, Absorption and Urease Activity................	51
	1. Experiment V. Reduction of foliar injury................	51
	2. Experiment VI. Absorption of urea.........................	60
	3. Experiment VII. Urease activity..	66
IV.	DISCUSSION....................................	73
	A. Response of Tomatoes to Urea Sprays.....	73
	B. Foliar Injury, Absorption and Urease Activity............................	75
V.	SUMMARY AND CONCLUSIONS.......................	80
VI.	LITERATURE CITED..............................	82
VII.	APPENDIX......................................	85
VIII.	BIOGRAPHICAL ITEMS............................	95

INTRODUCTION

With the report in 1943 (13) that apple trees could absorb and effectively assimilate nitrogen applied in spray solution to the leaves, a new field in plant nutrition was opened. In general, nutritional sprays had been restricted to application of the minor elements. This report suggested the possibility of supplying plants with at least one of the major elements by spraying the foliage with solutions containing urea.

The sandy soils of Florida with their low exchange capacities present many problems in the maintenance of a level of fertility sufficient for good plant growth. Initially low in clay and organic matter, the soil is depleted of its nutrients by leaching during the rainy seasons. If only to supply a part of the major nutritional requirements of the plant, the introduction of foliar sprays offers promise of a partial solution to the problem.

A review of the literature in 1950 revealed that surprisingly few investigations had been conducted with urea. Nevertheless, it was released to the farmer with suggestions for use on a wide variety of fruit and vegetable crops. It was evident that further investigations were necessary.

The present study, therefore, was undertaken primarily to evaluate the practice of nitrogen fertilization by the use of foliar sprays of urea. Also, it was hoped that information could be gained on some of the factors involved in the absorption and assimilation of urea. Since the tomato is of greatest economic importance in the Florida vegetable industry and since the plant offers many characteristics desired in this type of investigation, it was selected for this study.

REVIEW OF LITERATURE

The correction of certain deficiencies by spraying with solutions containing the necessary micro-nutrients has led to attempts to improve growth and vigor of many plants by foliar fertilization with nitrogen, phosphorus, and potassium. A great interest has developed in this field as the result of a report in 1943 by Hamilton, Palmiter, and Anderson (13), stating that urea sprays were beneficial to McIntosh apples grown in New York. Several nitrogen carriers were tested along with urea. Nitrate sprays proved to be quite injurious to the foliage. Ammonium sulfate did not injure the foliage nor did it show any beneficial effect. Solutions containing 10 pounds of urea per 100 gallons of water caused some burning of the leaves. Trees receiving urea solutions at concentrations of 5 pounds per 100 gallons of water were not injured. Analysis of the leaves showed that the nitrogen content had increased appreciably over that of unsprayed trees.

Foliar fertilization of apples with urea sprays has been investigated by a succession of workers at the New York stations. Fisher, Boynton, and Skodvin (7) in 1948 reported results of field trials in which McIntosh apples had been sprayed with Nu Green (urea) at concentrations of 5 pounds per 100 gallons of water. In a year which they considered unfavorable for fruit set in three western New

York orchards, 3 pounds of urea applied to the soil in the early spring were more effective in increasing terminal growth, set, and yield of fruit than the same amount applied in three sprays commencing at the petal-fall stage. They observed that spray applications resulted in slightly more fruit color than the soil applications.

Fisher and Cook (8) reported further results obtained in 1948 and 1949 with McIntosh apples. Four different levels of soil nitrogen applications were compared with a three-spray treatment and a four-spray treatment. They found that both spray treatments resulted in nitrogen and chlorophyll levels which were greater in June than when an equal amount of nitrogen was applied to the soil. At harvest time nitrogen and chlorophyll levels for all treatments were about equal. The three-spray treatment resulted in greater yield than soil applications of similar poundage.

Cook (5) cited the work of Harley, Moon, and Regeimbal (14) who found that leaves of York Imperial apples were able to absorb urea from spray applications. They applied Nu Green in concentrations of 5 pounds per 100 gallons of water to individual branches when they were in full bloom and then again fifteen days later. A cover was placed on the ground to prevent any urea from reaching the soil, where it could be absorbed by the roots. Forty-nine days later they noted that the leaves of sprayed branches were darker green in color than those of unsprayed branches.

Statistical analysis of the chemical data showed that sprayed leaves were significantly higher in chlorophyll and total nitrogen at the .05 level than unsprayed leaves.

Proebsting (19) observed that the leaves of apples and figs sprayed with urea were darker green in color than those in the control treatment. He also found that urea increased the yield of olive trees.

Merrill (18) reported that the yield from peach trees sprayed with urea was significantly higher at the .01 level as compared with the yield from unsprayed trees.

The possibility that nitrogen from nitrogenous fungicides might be assimilated by plants was investigated by Wentzler and White (24). They made three applications of Fermate, Crag 341, Parathion, and Phenothiozine to fully expanded leaves of young McIntosh and Stayman Winesap potted apple trees. They observed no significant changes in chlorophyll levels as compared with leaves of check trees ten days after spraying. Nu Green was added to the fungicide in other tests. Nu Green combined with Crag 341 spray resulted in a greater chlorophyll content per unit area of leaf surface than Nu Green combined with sulfur.

Other fruit crops have shown varying response to urea sprays. Although Merrill reported that peaches responded to urea sprays, other investigators have reported negative results with the same crop. Weinberger, Prince, and

Havis (23) investigating the use of urea sprays on peach trees at Fort Valley, Georgia, found that leaf nitrogen at harvest time, yield, and terminal growth were unaffected. Observations throughout the growing seasons failed to show any change in leaf color of sprayed trees. At Beltsville, Maryland, the same workers obtained no significant difference in leaf color, fruit color, and ripening dates even though concentrations as high as 25 pounds of urea per 100 gallons of water were used.

Proebsting (19) found that several of the stone fruits failed to respond to urea sprays. They included sweet cherry, apricot, peach, and almond.

Norton (18) noted that Elberta peaches grown in sand cultures showed no early response to urea sprays. He detected a darker green leaf color late in the season. The following year he added hydrated lime and a suitable spreader to the urea solutions. Increase in the green coloring of the leaves was attributed largely to the addition of the two compounds.

In 1949 Jones and Parker (16) reported results obtained with urea sprays on citrus. Although the first sprays in 1947 were not successful, those applied in 1948 and 1949 resulted in increased leaf nitrogen ten to fifteen days after application.

The response of grapes to urea has been found by two groups of investigators to be negative. Mack and

Shaulis (17) observed that the leaves from grapes receiving applications of urea at 4, 8, and 12 pounds per 100 gallons of water were progressively greener as the concentration increased; but, when the leaves were analyzed for total nitrogen, no significant differences were shown.

Tests by Fleming and Alderfer (9) over a three-year period with urea sprays on Concord grapes failed to show any beneficial effects. Analysis of leaf tissue in 1946 showed no significant differences in leaf nitrogen. The authors pointed out that the nitrogen of the Concord grapes was maintained at a high level throughout the growing season by heavy applications of nitrogen to the soil.

Urea sprays on vegetable crops have received comparatively little attention. Wittwer (18) reported that urea sprays of 3 to 4 pounds per 100 gallons of water for cucumbers, 10 to 12 pounds per 100 gallons of water for celery, and 5 to 6 pounds per 100 gallons of water for tomatoes proved to be "highly beneficial."

Hester (13) investigated the use of foliar sprays of urea as a means of supplying nitrogen to carrots and tomatoes. He obtained a 27 per cent increase in the yield of carrots by using urea sprays in a concentration of 20 pounds per 100 gallons of water. The yield of tomatoes was not significantly increased by applications of urea sprays as compared with equal amounts of nitrogen applied to the soil. Urea sprayed on young tomatoes grown for transplanting gave a 167 per cent increase in dry weight of young tomato plants as compared with applications of no nitrogen and a 98 per cent increase over "normal fertilizer treatment."

Danielson (18) applied urea sprays to a variety of vegetable crops at concentrations 2.5, 5.0, and 7.5 pounds of nitrogen per acre. Bountiful snap beans sprayed one week prior to first harvest were not affected in either yield or appearance. Radishes sprayed ten days before harvest showed some greening of the foilage in all rates, but the desired top growth did not occur. Old Dominion Blight-Resistant spinach, sprayed during January when tissue tests showed the plants to be deficient in nitrogen, was not responsive.

Danielson also tested the use of urea combined with potassium and phosphorus in a spray solution with a 1-2-1 ratio. The pH of the solution was adjusted to 6.0 with sodium hydroxide. Irish Cobbler potatoes showed no response when sprayed during a very dry period one month prior to harvest at rates equivalent to 5 and 10 pounds of 13-26-13 per acre. Rutgers tomatoes were sprayed over the foliage at weekly intervals with a balanced fertilizer solution. The total amount applied was 5 pounds of 13-26-13 per acre. A similar amount was applied directly to the soil in other plots. Both treatments appeared slightly better than unfertilized controls during early part of growing season, but these differences disappeared as the crop advanced. Yields were not significantly different. Black Valentine snap beans and Fordhook 242 lima beans, sprayed at weekly intervals with the equivalent of 5 pounds of 13-26-13 per acre beginning at emergence and continuing through harvest, were not improved in yield or color over control plots.

Foliar Injury

Several references have been made to leaf injury caused by urea sprays. Plants vary considerably in their ability to withstand a given concentration of urea in solution. Whereas, the tomato plant is injured under certain conditions when sprayed with 5 pounds of urea per 100 gallons of water, the carrot plant is not damaged with urea sprays four times more concentrated. The fact that urea cannot be used at high concentration with all crops makes it necessary to use unduly large amounts of water to apply a given quantity of the material.

In evaluating urea foliar sprays, some workers have attempted to reduce urea leaf injury by adding other compounds. Haas (12) found that nitrogen-deficient lemon cuttings were injured by spray solutions containing 44.8 pounds of urea per 100 gallons of water. When 2.5 pounds hydrated lime were added to the same amount of urea in solution for the second spray application to the cuttings, no leaf injury resulted.

Emmert and Klinker (6) reported that tomato plants grown under glass were injured by sprays containing 5 pounds of urea per 100 gallons of water. They found that they could increase the urea concentration tenfold without injury to the leaves by the use of a spray solution of urea and sucrose mixed in equal molar concentrations.

Mack and Shaulis (17) noted injury on Concord grape leaves when they were sprayed with 4 pounds of urea per 100 gallons. When the same amount of urea was added to a 3-3-100 Bordeaux mixture, the injury was prevented.

Absorption of Urea

A detailed study on the factors affecting the absorption of urea was made by Cook (5). He determined the rate of absorption by measuring the difference between a measured amount of solution atomized onto the surfaces of tagged apple leaves and the amount which after given time intervals could be washed off.

Briefly summarized, the following are some observations made by Cook in his studies:

1. Lower surfaces of apple leaves absorbed urea more quickly than the upper leaf surfaces. Similar results were obtained in limited trials with peach, plum, and sour cherry leaves.

2. There was a large variation in absorption from day to day and among individual trees on the same day.

3. High absorption was generally associated with high original leaf nitrogen level.

4. Light treatments, varied so as to affect carbohydrate level, had no influence on rate of absorption of urea.

5. The addition of sucrose to urea sprays reduced burning by decreasing the rate of urea uptake by the leaves.

6. Wilting of the leaves did not significantly alter the rate of absorption.

7. Rate of absorption was more than doubled by the addition of a wetting agent.

8. Calcium ions reduced absorption of urea slightly.

9. Variations in urea concentration did not affect percentage of absorption.

10. Half of the urea sprayed onto the leaves was absorbed in two to four hours, two-thirds in eight hours, and up to nine-tenths in forty-eight hours.

Assimilation of Urea

Very little is known about the metabolic assimilation of urea. The work of Reifer and Melville (20) suggests that the urea molecule is assimilated into a more complex compound. They supplied urea and ammonia in nutrient solutions to rye grass. The original traces of urea in leaves were increased thirty-five-fold in sixteen to twenty hours after urea was applied. Analysis showed consistent differences in glutamine values for plants receiving the two sources of nitrogen. They suggested that the urea might be used without conversion to ammonia. They reasoned that if urea were converted to ammonia no differences in glutamine values would have been obtained.

Urea can be converted rapidly to ammonia by the enzyme urease (22). There are conflicting reports regarding the presence of urease in the leaves of higher plants. Yamaguchi (25) found very small amounts of urease in the embryo and scutellum but none in other tissues of maize seedlings. Granick, on the other hand, detected the presence of measurable quantities of urease in all tissues of the soybean (10) and jack bean (11).

EXPERIMENTAL

The experimental data presented herein are divided into two parts. The first part, entitled Response of Tomatoes to Urea Sprays, consists of field and greenhouse tests designed to determine the value of foliar spray of urea in supplying nitrogen to tomatoes. The second part consists of three greenhouse and laboratory studies of problems of a physiological nature, which are closely interrelated with the field response studies. They are discussed under the general heading of Foliar Injury, Absorption, and Urease Activity.

A. Response of Tomatoes to Urea Sprays

Little information was available at the beginning of this study on the nitrogen nutrition of tomatoes with foliar sprays of urea. A preliminary test was conducted in the fall of 1950 to obtain as much general information as possible. When the results of the first experiment had been evaluated, the information was used to plan further studies. Two other field tests in the spring of 1951 and 1952 were designed to study foliar fertilization as a method of supplying supplemental nitrogen to tomato plants. The fourth study discussed in this section was conducted under closely controlled greenhouse conditions in an attempt to determine the degree of response which could be attributed to foliar absorption.

Experiment I (Fall, 1950)

Field test plots of tomatoes were grown in the fall of 1950 and sprayed with varying levels of Nu Green (urea). This study was undertaken to obtain general information on as many factors as possible which might be involved in the nitrogen nutrition of tomatoes with foliar sprays of urea.

Materials and methods

A 3 x 3 factorial design with four replications was used. Individual plots were thirty feet long and eight feet wide and contained two rows with ten tomato plants per row. Each replicate contained all possible combinations of three levels (0, 40, and 80 pounds per acre) of nitrogen applied to the soil with three similar levels of nitrogen in spray. The soil application consisted of a mixture of two parts nitrate, one part ammoniacal, and one part organic nitrogen. It was applied to the soil with the balance of the fertilizer materials. Nu Green, a completely water-soluble, synthetic urea compound containing 44 per cent nitrogen, was used as the source of spray nitrogen. It was sprayed on the tomato plants in concentrations of 2.5 pounds per 100 gallons of water.

All plots received applications of P_2O_5 (18 per cent superphosphate) at a rate of 84 pounds per acre and K_2O (potassium sulfate) at a rate of 60 pounds per acre.

Soil preparation was started in early August.

Repeated disking and harrowing were necessary to incorporate vegetative matter with the soil. Decomposition of the plant residues was not complete at planting time because of the lack of rain during this period.

Fertilizer, applied to the soil ten days before seeding, was placed in a band 6 inches below the surface of the soil. The soil was then fumigated with Dowfume W-40 by injecting 2.5 milliliters at each hill. Immediately following fumigation, the surface of the soil was watered by an overhead irrigation system.

Seed of the Jefferson variety of tomatoes was planted on August 28, 1950. It was placed by hand in hills thirty-six inches apart and covered with one-half inch of soil. When the plants had attained a height of approximately three inches, each hill was thinned to one plant. Early growth of the plants was retarded by heavy rains which fell in the first week of September.

The crop was cultivated intensively for weed control. When necessary water was supplied by overhead irrigation.

On September 19, 1950, when the leaf area of the young tomato plants was large enough to retain a major proportion of the material, the spray schedule was started. A self-propelled power spray machine was used to apply the material. Six applications, made at intervals of eight to ten days, were required to spray the designated amount of material at the concentration used.

In the course of the study, four leaf samples were collected from each plot. No spray material had been applied when the first sample was taken at the time of thinning of the plants. Additional samples were collected two days after the second, fourth, and last spray application. Uniformity in sampling was established by taking only the apical leaflet of the second and third complete leaves from the top of each plant. Immediately after representative leaf samples from each plot were collected, they were taken to the laboratory and washed three times in tap water to remove any foreign matter and urea adhering to the surfaces. After thorough drying in a force-draft oven at 70°C., the samples were ground in a Wiley mill to pass through a forty-mesh screen. Samples were stored in tightly closed glass containers until analyses could be made. The material was dried under 25 pounds of vacuum at 70°C. just before the analysis was made. Total nitrogen was determined by the technique described in A. O. A. C. official methods (1). It was adapted for micro-determinations by using proportionately smaller quantities of reagents and samples. All samples were analyzed in duplicates.

To compare the response of tomatoes in the early stages of growth to soil and foliar applications of nitrogen, the height of the plants was measured at the start of the spray schedule on September 22, 1950, and again after the second application on October 14, 1950.

The tomato plants were killed by heavy frost on November 22, 1950. Nevertheless, records were taken on total weight and number of fruits set from ten randomly selected plants. In addition, the ten plants selected were cut and stored in a barn for three weeks to obtain the air-dry weight of plant tissue.

Results

The first spray application of Nu Green was made on September 19, 1950, when the plants were approximately three inches in height. A study was made to determine the response of tomato plants in the early stages of growth to nitrogen foliar sprays. Average increases in height of tomato plants from September 22, to October 14, 1950, for the different treatments are given in Table 1. Significant increases at the .01 level were shown between levels of soil and leaf applications alike. Treatments with 40 pounds and with 80 pounds of nitrogen per acre did not differ significantly in either soil or foliar methods of application. Increases in height of plants in plots receiving 40 and 80 pounds of nitrogen per acre were significantly higher at the .01 level than increases in plants receiving no nitrogen.

Some interesting comparisons can be made from the individual combinations of nitrogen applications in Table 1.

Table 1

Increase in Height of Tomato Plants (cm.) from September 22, to October 14. Two Methods of Nitrogen Application; 3 Levels Each, 1950

Levels of Soil N Per Acre	Levels of Spray N Per Acre None	40 Lbs.	80 Lbs.	Mean For Levels of Soil N
None	13.43	16.43	17.25	15.70
40 lbs.	19.20	21.35	21.28	20.61
80 lbs.	21.30	21.20	20.50	21.00
Mean for Levels of Spray N	17.98	19.66	19.68	

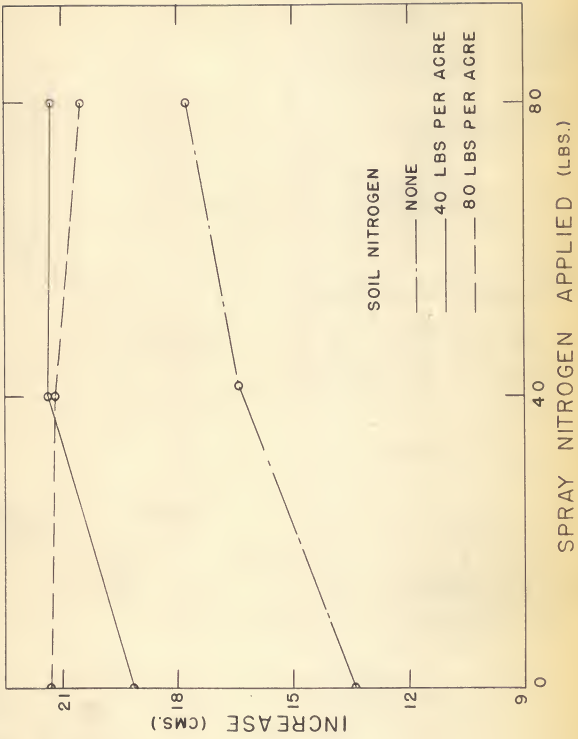
L.S.D. for Soil Treatment Means	.05 = 1.08 .01 = 1.47
L.S.D. for Leaf Treatment Means	.05 = 1.08 .01 = 1.47
L.S.D. for Individual Combination Means	.05 = 1.87 .01 = 2.54

Applications of 40 pounds of nitrogen per acre to the soil resulted in an average increase in height of tomato plants which was significantly higher at the .01 level than the same amount applied by foliar sprays. Similar results were shown by the eighty-pound levels of nitrogen.

Interaction, significant at the .05 level, was shown between levels of soil nitrogen and levels of leaf nitrogen. It is given graphically in Figure I. Plants in plots receiving 0 and 40 pounds of soil nitrogen showed a significant

Figure I

Increase in Height of Tomato Plants (cm.) from September to October 14. Two Methods of Nitrogen Application, 3 Levels Each, 1950.


INCREASE (CMS.)
9
12
15
18
21
SPRAY NITROGEN APPLIED (LBS.)
0
40
80
SOIL NITROGEN
NONE
40 LBS PER ACRE
80 LBS PER ACRE

increase in average height from foliar nitrogen sprays. On the other hand, plants in plots receiving 80 pounds of soil nitrogen were not significantly responsive to the foliar sprays of nitrogen.

The total nitrogen level of the leaves of tomato plants was determined on four dates in the course of the study (Table 2). Plants in plots receiving 40 and 80 pounds of nitrogen per acre from either soil or leaf applications were significantly higher at the .01 level than their corresponding control which received no nitrogen. It can be noted from Table 2 that the average total nitrogen content of the leaves of plants was almost equal within similar levels of soil and leaf nitrogen application. Each increase in amount of nitrogen from either soil or foliar applications gave highly significant increases in the nitrogen level of the leaves.

The first order interaction between soil and leaf applications was significant at the .01 level. Response from 40 pounds of nitrogen by foliar sprays was more evident, as measured by the nitrogen level in plants, in those plots which were initially low in nitrogen. It is shown graphically in Figure 2.

The average total nitrogen content of the tomato plants was significantly lower at the .01 level on October 26, 1950, (the third date of sampling) than on any of the

Table 2

Total Nitrogen (Per Cent Dry Weight Basis) of Tomato Leaf Tissue Sampled on Four Dates. Two Methods of Nitrogen Application, 3 Levels Each, 1950

Levels of Soil N Per Acre	Levels of Spray N Per Acre			Mean For Levels of Soil N
	None	40 Lbs.	80 Lbs.	
None	3.62	4.28	4.72	4.21
40 lbs.	4.18	4.72	5.08	4.66
80 lbs.	4.75	4.94	5.27	4.99
Mean for Levels of Spray N	4.19	4.65	5.03	

L.S.D. for Soil Treatment Means .01 = 0.17

L.S.D. for Spray Treatment Means .01 = 0.17

L.S.D. for Individual Combination Means .05 = 0.24
.01 = 0.32

three other dates (Table 3). Date of sampling is used here to designate the interval of time elapsing between planting and sampling.

Figure 3 illustrates the first order interaction between dates of sampling and levels of soil nitrogen which is significant at the .01 level. On the first date of sampling the differences in the nitrogen levels of the plant are large as a direct effect of the amounts of nitrogen applied to the soil initially. When the plants were sampled for the second time, two applications of spray nitrogen had

Figure 2

Total Nitrogen (Per Cent Dry Weight Basis) of Tomato Leaf Tissue Sampled on Four Dates. Two Methods of Nitrogen Application, 3 Levels Each, 1950.

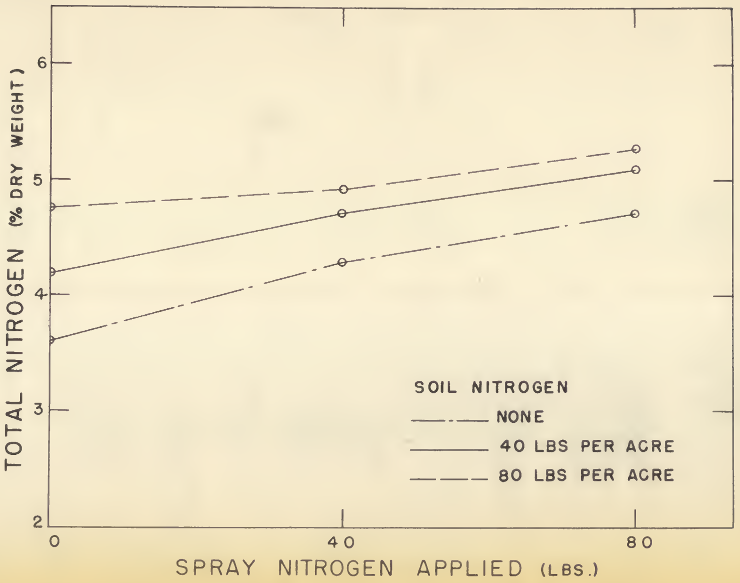

TOTAL NITROGEN (% DRY WEIGHT)
6
5
4
3
2
0
40
80
SPRAY NITROGEN APPLIED (LBS.)
SOIL NITROGEN
NONE
40 LBS PER ACRE
80 LBS PER ACRE

Table 3

Effect of Dates of Sampling on the Average Total Nitrogen Content of Leaves (Per Cent Dry Weight Basis) of Tomato Plants Receiving 3 Levels of Nitrogen Applied to the Soil before Planting, 1950

Levels of Soil N Per Acre	Dates of Sampling			
	Sept. 18	Oct. 3	Oct. 26	Nov. 9
None	4.14	4.51	3.19	5.00
40 lbs.	5.69	4.76	3.28	4.92
80 lbs.	5.97	5.30	3.49	5.22
Mean For Dates of Sampling	5.27	4.86	3.32	5.05

L.S.D. for Dates of Sampling Means .01 = 0.16

L.S.D. for Individual Combination Means .05 = 0.21
.01 = 0.28

been made. The effect of spray application of urea was evident by the increase shown in the nitrogen level of the plants in plots receiving no nitrogen at planting time. Plants in plots receiving 40 and 80 pounds of nitrogen per acre showed actual decreases in leaf nitrogen levels during the same period.

The average total nitrogen content of tomato plants sprayed with three levels of urea nitrogen was influenced by the date of sampling (Table 4). The interaction significant at the .01 level is shown graphically in Figure 4. It can be noted that on October 23, 1950, after only two applications

Figure 3

Effect of Dates of Sampling on the Total Nitrogen Content of Leaves of Tomato Plants Receiving 3 Levels of Nitrogen Applied to the Soil before Planting, 1950.

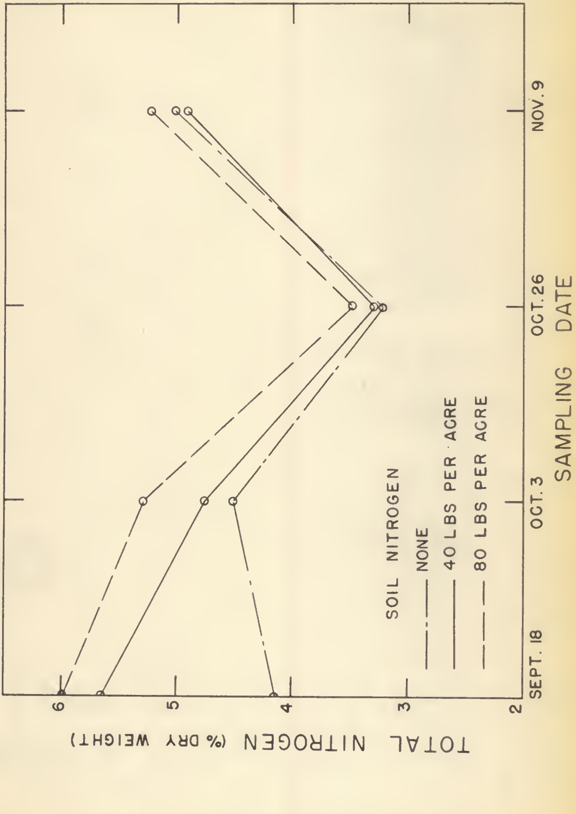

TOTAL NITROGEN (% DRY WEIGHT)
6
5
4
3
2
SEPT. 18
OCT. 3
OCT. 26
NOV. 9
SAMPLING DATE
SOIL NITROGEN
NONE
40 LBS PER ACRE
80 LBS PER ACRE

Table 4

Effect of Dates of Sampling on the Total Nitrogen Content of Leaves (Per Cent Dry Weight Basis) of Tomato Plants Receiving Three Levels of Spray Nitrogen, 1950

Levels of Spray N Per Acre	Dates of Sampling			
	Sept. 18	Oct. 3	Oct. 26	Nov. 9
None	5.47	4.39	2.90	3.99
40 lbs.	5.27	4.96	3.37	4.99
80 lbs.	5.06	5.20	3.69	6.15
Mean for Dates of Sampling	5.27	4.85	3.32	5.05

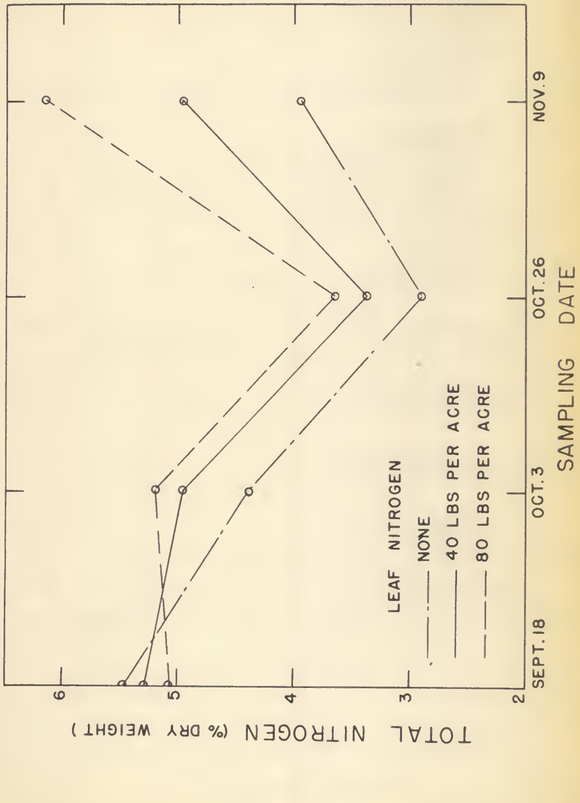
L.S.D. for Individual Combination Means .05 = 0.21
.01 = 0.28

of spray nitrogen, plants in plots receiving spray nitrogen were higher in total nitrogen than the controls. This relationship was more or less constant throughout the growth period of the tomato plants.

The fruit had not matured when the plants were killed by frost on November 22, 1950. Nevertheless, it was harvested and a statistical analysis was made on yields obtained. Only 50 per cent of the plants, randomly selected from each plot, were used for studies of total weight of fruit harvested, number of fruits set, and total dry matter produced.

Figure 4

Effect of Dates of Sampling on the Total Nitrogen Content of Leaves of Tomato Plants Receiving 3 Levels of Spray Nitrogen, 1950.


TOTAL NITROGEN (% DRY WEIGHT)
6
5
4
3
2
SEPT. 18
OCT. 3
OCT. 26
NOV. 9
SAMPLING DATE
LEAF NITROGEN
NONE
40 LBS PER ACRE
80 LBS PER ACRE

Average weight of the immature fruit harvested from one-half of the tomato plants in each plot is given in Table 5. Applications of 40 and 80 pounds of soil nitrogen to the acre resulted in increases in weight of fruit harvested which were significantly higher at the .01 level than the control treatment.

Table 5

Weight (gm per Plot) of Immature Tomato Fruit Harvested from 50 per cent of the Plants. Two Methods of Nitrogen Application, 3 Levels Each, 1950

Levels of Soil N Per Acre	Levels of Spray N Per Acre			Mean For Levels of Soil N
	None	40 Lbs.	80 Lbs.	
None	538	1,440	1,496	1,156
40 Lbs.	1,582	2,564	2,512	2,219**
80 Lbs.	2,317	2,369	1,760	2,149**
Mean for Levels of Spray N	1,479	2,124*	1,923	

L.S.D. for Soil Treatment Means	.05 = 471 .01 = 638
L.S.D. for Spray Treatment Means	.05 = 471 .01 = N.S.
L.S.D. for Individual Combination Means	.05 = 747 .01 = 1,012

In comparisons of leaf spray applications, statistical significance was shown at the .05 level. Application of

40 pounds of spray nitrogen per acre resulted in a significant increase in weight of fruit harvested over the control treatment. An increase was not shown by the eighty pound spray treatment.

In a comparison of individual combinations of nitrogen treatments in Table 4, it can be seen that the weight of immature tomatoes harvested in plots receiving only 40 pounds of leaf nitrogen did not differ significantly from plots receiving comparable amounts in the soil. Applications of 80 pounds of nitrogen per acre to the soil resulted in significant increases at the .05 level in weight of fruit over plots receiving equal amounts by spray application alone.

Analysis of the data on weight of fruit showed that the interaction between soil and leaf applications was not statistically significant.

The average number of fruits set per plot was increased by the application of nitrogen to the soil as well as to the foliage (Table 6). Whereas the forty-pound spray application increased fruit-set significantly at the .01 level, the increase from the eighty-pound spray treatment was significant at the .05 level. From these results it can be seen that, by increasing the amount of nitrogen, the number of fruits set was decreased slightly.

Comparisons among individual combinations

Table 6

Number of Fruits Harvested per Plot (from 50 Per Cent of the Plants). Two Methods of Nitrogen Application, 3 Levels Each, 1950

Levels of Soil N Per Acre	Levels of Spray N Per Acre			Mean For Levels of Soil N
	None	40 Lbs.	80 Lbs.	
None	20.0	39.8	32.0	30.6
40 lbs.	28.3	55.0	54.3	45.9^{**}
80 lbs.	45.3	47.0	39.3	43.9^{**}
Mean for Levels of Spray N	31.2	47.2^{**}	42.5^{*}	

L.S.D. for Soil Treatment Means .05 = 8.8
.01 = 12.0

L.S.D. for Spray Treatment Means .01 = 8.8
.05 = 12.0

L.S.D. for Individual Combination Means .05 = 15.3
.01 = 20.7

(Table 6) showed that 40 pounds of spray nitrogen significantly increased (.01 level) the number of fruits set over the control. The same level applied to the soil, however, did not show a significant increase. The addition of spray nitrogen generally increased fruit set except in those cases where the soil nitrogen was initially high.

Statistical analysis of the data on the number of fruits set showed that the interaction between soil and spray applications was not significant.

A comparison of the average air dry weight of tissue produced from 50 per cent of the plants in each plot is shown in Table 7. Significant differences were shown by soil and spray applications alike. The increases resulting from spray applications were larger than increases from soil nitrogen.

Table 7

Air Dry Weight (gm per Plot) of Plant Tissue Harvested (50 Per Cent of the Plants). Two Methods of Nitrogen Application, 3 Levels Each, 1950

Levels of Soil N Per Acre	Levels of Spray N Per Acre			Mean For Levels of Soil N
	None	40 lbs.	80 lbs.	
None	296	740	927	654
40 lbs.	362	839	1,059	753
80 lbs.	719	1,080	1,088	962**
Mean for Levels of Spray N	459	886**	1,025**	

L.S.D. for Soil Treatment Means .05 = 172
.01 = 232

L.S.D. for Spray Treatment Means .05 = 172
.01 = 232

L.S.D. for Individual Combination Means .05 = 297
.01 = 403

The interaction between the two methods of nitrogen applications was not statistically significant. It can be noted from Table 7 that increasing the amount of nitrogen by either method of application increased the amount of dry matter produced. The dry matter produced in plots receiving 40 pounds of spray nitrogen was significantly higher at the .05 level than that produced in plots receiving 40 pounds of soil nitrogen.

Experiment II (Spring, 1951)

In preliminary studies in the fall of 1950, it was found that the application of urea nitrogen to the foliage of tomatoes was not so beneficial in the early stages of growth as nitrogen applied to the soil. In view of these results, this experiment was designed to determine the effectiveness of urea as a supplemental source of nitrogen as compared with sidedressings of equal amounts of nitrogen from sodium nitrate.

Materials and methods

A 2 x 2 factorial design with eight replications was used. Each replicate contained all possible combinations of two levels of soil nitrogen (20 and 40 pounds per acre) applied at transplanting time with sidedress or spray applications of additional nitrogen so that all plots received a total of 50 pounds per acre. Individual plots were thirty-six feet long and four feet wide and separated from one another by a single row of tomatoes. Plots contained twelve tomato plants spaced three feet apart.

The initial soil nitrogen was applied before transplanting in a mixture with the balance of the fertilizer materials. It was composed of equal parts of nitrate and ammoniacal nitrogen from sodium nitrate and ammonium sulfate respectively. Phosphorus and potassium were applied at the same rate as in Experiment I.

Materials used as supplemental nitrogen sources were sodium nitrate for sidedressing and Nu Green for spray applications. The latter was used in a concentration of 2.5 pounds per 100 gallons of water with Triton added as a spreader.

The soil was disked and harrowed several times prior to fumigation on March 6, 1951. Dowfume W-40 was applied at the rate of 15 gallons per acre by an automatic dispenser mounted on a tractor.

On March 15, 1951, the fertilizer was applied to the soil. It was placed four inches deep and four inches to each side of the center of the row.

Tomato seed of the Jefferson variety treated with Spergon was planted in flats in the greenhouse on February 10, 1951. The plants were transplanted in the field on March 20, 1951.

Application of supplemental nitrogen was started on May 7, 1951, when the fruit had begun to set. Plots which were to receive sidedressings of sodium nitrate received one-half of the total amount on that date. Similarly, plots which were to receive 10 pounds of spray nitrogen received one-half of that amount at that time. Since it was impossible to apply one-half of the nitrogen in one spray operation in plots receiving 30 pounds by that method, three applications at intervals of about five days were made. When all plots had received 50 per cent of the supplemental nitrogen, application of the remaining 50 per cent was carried out in a similar fashion. The last spray application was made May 31, 1951.

Nitrate nitrogen determinations on mature leaf petioles of the plants in all treatments were made on May 16, 1951, when 50 per cent of the supplemental nitrogen had been applied. Other nitrate determinations were made on June 7, 1951, after plots had received all of the supplemental nitrogen. Tomato plants from each test plot were sampled by taking mature leaf petioles. Analysis was made on fresh tissue. It was necessary to sample a few plots at a time and to make the analysis as soon as possible after collection. The technique was adapted from a method used to determine nitrates in soils (15). Instead of shaking the mixture as is recommended for soils, the plant tissue was thoroughly macerated in a Waring blendor before adding the reagents and then mixed again before filtering.

Leaf samples for total nitrogen determinations were collected on May 22 and June 11, 1951. Samples were handled by the procedure discussed in Experiment I.

"Mature-green" fruits were harvested five times starting June 4 and ending June 25, 1951. Tomatoes were graded into three classes according to quality. Records were kept on total weight and number of fruits harvested on each date.

Results

Analysis of the results given in Table 8 showed that initial level of soil nitrogen and sources of supplemental nitrogen did not significantly affect yield of tomatoes as measured by total weight and number of fruits harvested.

Table 8

Effect of Split-applications of Nitrogen on Yield of Tomatoes, 1951

Type of Split-application of Nitrogen		Total Weight (Pounds)	Total Number Fruits
20 lbs. soil	30 lbs. sidedress	598.3	2,052
20 lbs. soil	30 lbs. spray	675.7	2,328
40 lbs. soil	10 lbs. sidedress	675.3	2,236
40 lbs. soil	10 lbs. spray	698.2	2,305

No significant differences.

It can be noted in Table 8 that plots receiving split-applications of 20 pounds of nitrogen in the soil prior to transplanting plus 30 pounds of sidedress nitrogen were lower in yields than all the remaining plots. A breakdown of the sum of squares showed that those plots were significantly lower at the .05 level in weight (Table 9) and number of fruits (Table 10) than plots of the other three treatments.

Table 9

Analysis of Variance: Effect of Split-applications of Nitrogen on Total Weight of Tomatoes, 1951

Source of Variance	Degrees of Freedom	Variance	F Value
No. 1 vs. Nos. 2, 3, & 4	1	637.7	4.60*
No. 2 vs. No. 3 vs. No. 4	2	21.5	-
Error	21	138.6	-

*Significant at .05 level.

Table 10

Analysis of Variance: Effect of Split-applications of Nitrogen on Total Number of Fruits Set, 1951

Source of Variance	Degrees of Freedom	Variance	F Value
No. 1 vs. 2, 3, & 4	1	5,295.0	4.32*
No. 2 vs. 3 vs. No. 4	2	286.5	-
Error	21	1,225.0	-

*Significant at .05 level.

To determine the effect of different types of split-applications of nitrogen on date of maturity of tomatoes, the harvest was divided into early and late periods. Statistical analysis of the data showed that treatments did not significantly affect date of maturity (Table 11).

A comparison of the average nitrate levels of plants in plots receiving four different types of split-applications

Table 11

Effect of Split-applications of Nitrogen
on Date of Maturity of
Tomatoes, 1951

Type of Split-application of Nitrogen		Weight of Tomatoes (lbs.) For Each Period	
		June 4 - June 14	June 15 - June 25
20 lbs. soil	30 lbs. sidedress	321.3	277.0
20 lbs. soil	30 lbs. spray	380.0	295.7
40 lbs. soil	10 lbs. sidedress	376.3	299.0
40 lbs. soil	10 lbs. spray	375.0	340.6

No significant differences.

Table 12

Effect of Date of Sampling on the Nitrate Levels of
Tomato Plants Receiving Four Types of Split-
applications of Nitrogen, 1951

Type of Split-application of Nitrogen		Average Nitrate Nitrogen (PPM)	
		May 16, 1951	June 7, 1951
20 lbs. soil	30 lbs. sidedress	701	601
20 lbs. soil	30 lbs. spray	664	245
40 lbs. soil	10 lbs. sidedress	1014	765
40 lbs. soil	10 lbs. spray	1091	269

of nitrogen is given in Table 12. The results showed that on May 16, the average nitrate-nitrogen levels of the plants were more or less proportional to the quantity of nitrogen applied at transplanting time. The nitrate-nitrogen levels of the plant in plots receiving their supplemental nitrogen from urea sprays dropped considerably more on the second date of sampling than plants in plots receiving side-dressings of sodium nitrate.

Total nitrogen content of the plants was determined on two dates after the application of supplemental nitrogen was started. There were no significant effects of initial levels of soil nitrogen on the total nitrogen content of the plants. A significant difference was shown between sources of supplemental nitrogen. Plants from plots receiving side-dressings of sodium nitrate were significantly higher at the .05 level in average total nitrogen than plants from plots receiving urea sprays (Table 13).

The difference in average total nitrogen between dates of sampling, as well as the first order interaction between dates of sampling and sources of supplemental nitrogen, were highly significant (Table 13). The average total nitrogen content of plants from urea-sprayed plots dropped from 4.72 per cent on May 22, to 3.88 per cent on June 11. During the same period plants from sodium nitrate side-dressed plots dropped in percentage nitrogen from 4.75 to 4.20. The first order interaction is shown graphically in Figure 5.

Table 13

Effects of Sources of Supplemental Nitrogen and Dates of Sampling on the Average Total Nitrogen Content of Leaves (Per Cent Dry Weight Basis) of Tomato Plants, 1951

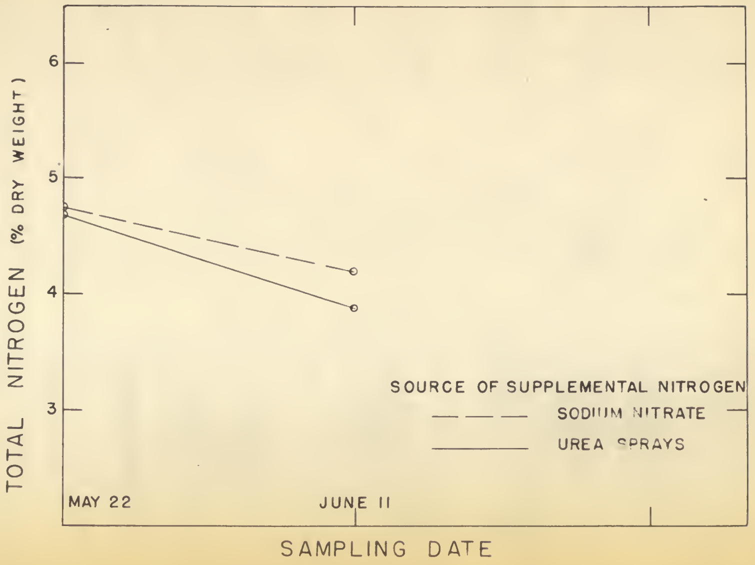
Sources of Supplemental Nitrogen	Dates of Sampling		Means for Sources
	May 22	June 11	
Sodium Nitrate Side-dressings	4.75	4.20	4.48*
Urea Sprays	4.72	3.80	4.30
Meansfor Dates	4.74**	4.04	

*Significant at .05 level.

**Significant at .01 level.

Figure 5

Effect of Sources of Nitrogen and Dates of Sampling
on the Total Nitrogen Content of Leaves of
Tomato Plants. Four Split-applications
of Nitrogen, 8 Replications, 1951.


TOTAL NITROGEN (% DRY WEIGHT)
6
5
4
3
MAY 22
JUNE 11
SAMPLING DATE
SOURCE OF SUPPLEMENTAL NITROGEN
SODIUM NITRATE
UREA SPRAYS

Experiment III (Spring, 1952)

In the spring of 1951 a test was conducted to compare urea sprays with sodium nitrate as supplemental sources of nitrogen for tomatoes. The results failed to show significant differences in dates of maturity, weights, or numbers of tomatoes harvested. Under the conditions of low rainfall which prevailed during the growing season, the nitrogen applied before transplanting was not subjected to excessive leaching. It was possible, therefore, that additional nitrogen was not required by the plants.

In the spring of 1952 another test was conducted to compare urea sprays with sodium nitrate as supplemental sources of nitrogen for tomatoes. The test was modified to include one treatment which would receive no additional nitrogen after transplanting.

Materials and methods

Four supplemental nitrogen treatments for tomatoes were tested in a randomized-block design with four replications. The plots, divided into two rows with ten plants per row, were 11.4 feet wide and 25 feet long. All plots were separated from one another by a single row of tomatoes.

Fumigation and fertilization, with the exception of nitrogen, were the same as in the spring of 1951. Twenty pounds of nitrogen from ammonium nitrate were applied to the soil with the balance of the fertilizer materials before transplanting.

Tomato plants of the Rutgers variety were started in flats in the greenhouse on February 13, 1952, and transferred to the field on March 24, 1952.

Supplemental nitrogen treatments were as follows: (1) no additional nitrogen; (2) sidedressings of 30 pounds of nitrogen per acre from sodium nitrate; (3) foliar spray of 30 pounds of nitrogen per acre from Nu Green; and (4) 30 pounds of nitrogen per acre from Nu Green sprayed on the ground around the plants. Application of supplemental nitrogen was started on May 10, 1952, and ended on June 2, 1952. Urea was applied in a concentration of 4 pounds per 100 gallons of water.

Tomatoes were harvested four times from June 6 to June 24, 1952. Records were kept on weight and number of fruits harvested from each plot.

Results

Urea sprays and sodium nitrate were compared as sources of supplemental nitrogen for tomatoes. Conditions prevailing during the growing season were not conducive to the best production of tomatoes. Prolonged cool weather after transplanting, with several heavy rains following, retarded the growth of the plants considerably.

The results given in Table 14 show that there were no significant differences among the four treatments in weight or numbers of tomatoes harvested.

Table 14

Effect of Supplemental Nitrogen Treatments on the Weight and Number of Tomatoes Produced, 1952

Supplemental Nitrogen Treatments	Total Weight (Pounds)	Total Number Fruits
None	120.4	499
20 lbs. NaNO3 Sidedress	116.4	474
20 lbs. Urea Foliar Spray	131.9	485
20 lbs. Urea Soil Spray	116.9	468

No significant differences.

Experiment IV (Greenhouse, 1951)

A large proportion of the urea solution sprayed on tomatoes under field conditions probably reaches the soil where it can be absorbed by the roots. This experiment was designed primarily to eliminate this factor and to study the response of tomatoes to urea from direct foliar absorption.

Materials and methods

Tomato plants started in flats were transplanted to eight-inch pots on January 30, 1951. The type of soil used was an Arredondo fine sand taken from the Agricultural Experiment Station Farm. Phosphorus and potassium were added to it at a rate equivalent to that used in the field experiment. Potted plants were placed on a greenhouse bench and allowed to grow for ten days before spray applications were started.

In a split-plot design with four replications, two concentrations (2.5 and 5.0 pounds of Nu Green per 100 gallons of water) were compared with a control treatment receiving no nitrogen except that which was initially present in the field soil. After completion of the spray schedule the plots in each of the above treatments were divided into three dates for sampling at weekly intervals. Thus, the thirty-six plots, each consisting of two pots, were divided so that twelve plots were sampled on each date. The plants had not reached maturity when they were sampled.

On February 9, 1951, the spray schedule was started. All plants were sprayed six times at weekly intervals. The last application was made on March 8, 1951. Throughout the experiment none of the material applied to the leaves was allowed to reach the roots. This was easily accomplished by placing covers over the soil during the spray operation. Similarly, extreme care was taken in watering the plants. Water was applied daily from a cup directly to the surface of the soil in each pot.

When the spray schedule was started, a measurement of the height of the tomato plants was taken. Another measurement was made on the date the plants were sampled. The fresh weight of the plants was recorded before leaf samples were taken for subsequent determination of total nitrogen contents. On the last date of sampling, a composite sample of the roots from all the plants in each treatment was taken and the total nitrogen content was determined.

Results

Considerable leaf-tip and marginal burn was noted on the plants receiving the higher concentration of 5.0 pounds of urea per 100 gallons of water. Solutions containing 2.5 pounds per 100 gallons of water resulted in no apparent injury to the plants.

Increase in height of tomato plants was used as a measure of the response to urea sprays. The results obtained on effect of varying concentrations of urea and dates of sampling on the average increase in height of tomato plants are given in Table 15. The average increase of height of tomato plants in plots receiving six applications of urea at 2.5 pounds per 100 gallons of water was significantly higher at the .05 level than that of the control plants. With the higher concentration of 5.0 pounds per 100 gallons of water, significant increases were shown at the .01 level.

Table 15

Effect of Spray Application of Urea and Dates of Sampling on the Increase in Height (cm.) of Tomato Plants, Greenhouse, 1951

Pounds of Urea Per 100 Gallons of Water	Dates of Sampling			Means For Urea Treatments
	March 10	March 17	March 24	
None	31.9	35.8	42.2	36.6
2.5	34.5	38.2	44.5	39.1*
5.0	36.6	41.4	47.2	41.7**
Means For Dates of Sampling	34.3	38.5**	44.6**	

L.S.D. for Urea Treatment Means .05 = 1.0
.01 = 2.9

L.S.D. for Dates of Sampling Means .05 = 2.8
.01 = 3.8

The average increase in height was significant at .01 level between dates of sampling (Table 15). The interaction between dates of sampling and spray treatments was not statistically significant.

Analysis of the data on average fresh weight of plants showed that there were significant increases in plots receiving urea sprays as compared with plots receiving no urea (Table 16). It can be noted that as the concentrations

Table 16

Effect of Spray Applications of Urea and Dates of Sampling on the Fresh Weight (gm.) of Tomato Plants, Greenhouse, 1951

Pounds of Urea Per 100 Gallons of Water	Dates of Sampling			Means For Urea Treatments
	March 10	March 17	March 24	
None	54.2	62.9	78.4	65.2
2.5	63.0	82.2	82.8	76.0*
5.0	72.9	81.0	103.3	85.7**
Means for Dates of Sampling	63.4	75.4**	88.2**	

L.S.D. for Urea Treatment Means .05 = 8.0
.01 = 12.1

L.S.D. for Dates of Sampling Means .05 = 8.3
.01 = 11.4

of the urea solutions increased, the fresh weight of plant tissue which was produced increased also.

There were significant differences between dates of sampling (Table 16). The interaction between urea treatments and dates of sampling was not statistically significant.

A comparison of average nitrogen levels of tomato plants is given in Table 17. Significant increases were shown at the .01 level in average total nitrogen levels of tomato plants from plots receiving spray applications of urea.

Table 17

Effect of Spray Applications of Urea and Dates of Sampling on the Total Nitrogen Content of Leaves (Per Cent Dry Weight) of Tomato Plants, Greenhouse, 1951

Pounds of Urea Per 100 Gallons of Water	Dates of Sampling			Means For Urea Treatments
	March 10	March 17	March 24	
None	1.34	1.47	1.29	1.37
2.5	1.91	1.65	1.60	1.72**
5.0	2.56	1.87	1.86	2.10**
Means For Dates of Sampling	1.94**	1.66	1.58	

L.S.D. for Urea Treatment Means .05 = 0.12
.01 = 0.16

L.S.D. for Dates of Sampling Means .05 = 0.09
.01 = 0.12

Highly significant differences were also noted between dates of harvest (Table 17). On March 10, 1951, the average per cent total nitrogen of all plants was 1.94 as compared with 1.66 on March 17 and 1.58 on March 24. The interaction between urea treatments and dates of sampling was significant at the .01 level. It is illustrated graphically in Figure 6. The total nitrogen level of the plants in plots receiving no urea sprays dropped proportionally less from March 10 to March 24, 1951.

The total nitrogen content of composite samples of roots of tomatoes from each treatment was determined on the last date of sampling. The results showed that two weeks after the last application of urea, the roots of plants in plots receiving urea sprays had a higher total nitrogen content than those of the control plants (Table 18). The number of observations made was too small to analyze statistically.

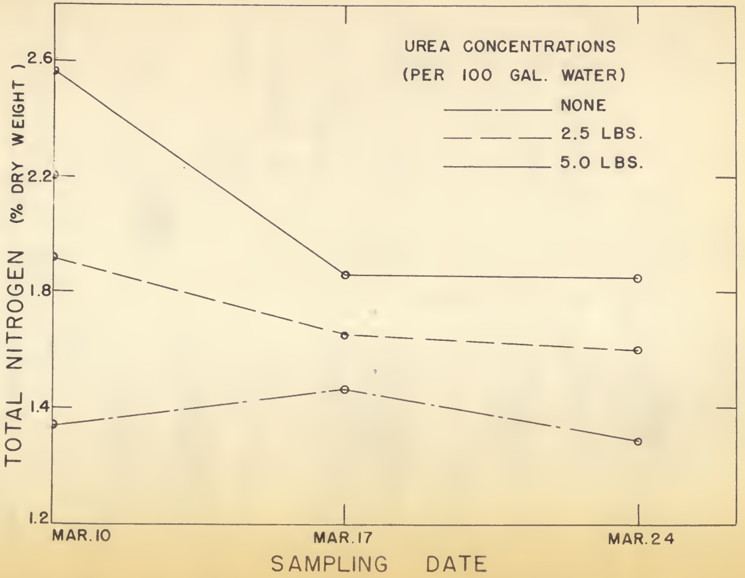
Table 18

Total Nitrogen Content of Roots (Per Cent Dry Weight) of Tomato Plants Two Weeks After Urea Spray Schedule was Completed, Greenhouse, 1951

Urea Concentration	Per Cent Nitrogen
None	1.09
2.5 lbs. per 100 gal. water	1.16
5.0 lbs. per 100 gal. water	1.28

Figure 6

Effect of Spray Applications of Urea and Dates of Sampling on the Total Nitrogen of Leaves of Tomato Plants, Greenhouse, 1951.


UREA CONCENTRATIONS
(PER 100 GAL. WATER)
NONE
2.5 LBS.
5.0 LBS.
TOTAL NITROGEN (% DRY WEIGHT)
2.6
2.2
1.8
1.4
1.2
MAR. 10
MAR. 17
MAR. 24
SAMPLING DATE

Studies on Foliar Injury, Absorption, and Urease Activity

In evaluating the response of tomato plants to urea sprays, numerous problems arose which warranted further study. More information was needed on urea sprays with reference to causes and reduction of foliar injury, rate of absorption, and metabolism of urea.

The three experiments reported herein are interrelated studies entitled: (1) Reduction of foliar injury; (2) Absorption of urea; and (3) Urease activity. The first study includes the results obtained in a search for materials which are capable of reducing foliar injury when added to urea solutions. The second study was conducted to learn, if possible, the mode of action involved in reduction of foliar injury. It was hoped that, by measuring the rate of absorption and disappearance of urea, something could be learned about reduction of foliar injury as well as the metabolism of urea. In the third study, measurements of urease activity of the tomato plant were made. This study was made to determine whether or not urea is hydrolyzed to any extent to ammonia and carbon dioxide by the tomato plant.

Experiment V Reduction of foliar injury

Investigators have drawn attention to the fact that urea may cause severe injury to the foliage of many plants. The use of foliar sprays of urea is often limited to solutions of relatively low concentrations. Bordeaux mixture, hydrated lime, and sucrose have been found to reduce foliar injuries to varying degrees when mixed with urea sprays. This investivation was undertaken in an attempt to find additional materials which, when added to urea solutions, would reduce injury to the leaves of tomatoes.

Materials and methods

Five tests were conducted under greenhouse conditions from August, 1950 to January, 1951. As each test was completed and the results evaluated, the information gained was used to eliminate those compounds which showed no promise and to study further those which indicated reduction of urea injury to tomato leaves. The materials used in each test are given together with the results. All chemicals were reagent grade materials.

Tomatoes of the Rutgers variety were used for this study. Plants for each test were started in flats and transferred to eight-inch pots. A 4-7-5 fertilizer at the rate of 1500 pounds per acre was added to the soil. When the plants attained a height of approximately sixteen inches, they were sprayed with the test solutions. Triton (a proprietary spreader compound) was added to the test solutions to reduce the formation of large water drops on the surface of tomato leaves. A one-liter sprayer with 60 pounds of pressure was used to deliver a very fine mist. No records were kept on temperature, relative humidity, or light conditions.

Results

Four to six days after the tomato plants had been sprayed for each test they were examined and a rating was made of the relative severity of the urea injury to the foliage of each plant. The plants were rated on a scale from 0, which indicates no injury, to 10, which indicates that the plants were injured beyond recovery. Each test solution was sprayed on three plants. The score is an average of the injury ratings of individual plants in each treatment.

Sucrose and calcium chloride were added in various concentrations to urea solutions in the first test. The results are given in Table 19. In accordance with the results reported by Emmert and Klinker (2), sucrose was found to reduce the extent of burning considerably. Plants sprayed with urea solutions containing calcium chloride were injured just as severely as those sprayed with urea solutions without calcium chloride. It was noted, however, that burning was somewhat delayed by the addition of calcium chloride.

Calcium chloride was selected for further study in the second test. Magnesium sulfate was tested in a limited number of solutions. From the results given in Table 20 it can be seen that Calcium Chloride was essentially ineffective in reducing urea injury to tomato foliage. When 0.18 mole of Magnesium Sulfate was added to 0.4 mole of urea in a liter

Table 19

Effect of Sucrose and Calcium Chloride on the Reduction of Urea Injury to Young Tomato Plants Grown in the Greenhouse (First Test)

Materials and Concentrations Used in Each Solution	Average Score of Urea Injury[a]
.05 M urea alone	0.0
.05 M urea + .05 M sucrose	0.0
.05 M urea + .02 M $CaCl_2$	0.0
.1 M urea alone	1.0
.1 M urea + .1 M sucrose	0.0
.1 M urea + .04 M $CaCl_2$	1.0
.2 M urea alone	3.0
.2 M urea + .2 M sucrose	1.0
.2 M urea + .08 M $CaCl_2$	3.0
.4 M urea alone	5.0
.4 M urea + .4 M sucrose	2.0
.4 M urea + .16 M $CaCl_2$	5.0
.8 M urea alone	9.0
.8 M urea + .8 M sucrose	4.0
.8 M urea + .32 M $CaCl_2$	9.0

[a] Based on a scale from 0, which indicates no injury, to 10, which indicates the plants were injured beyond recovery.

of solution, a noticeable reduction in leaf burn resulted as compared with 0.4 M solutions of urea alone.

Since magnesium sulfate showed promise as a material which might reduce injury to the foliage of tomatoes, it was studied in more detail in the third test. Other materials included in this test were calcium oxide, calcium acetate, magnesium chloride, and citrus molasses. None of the materials tested, except magnesium sulfate, reduced urea injury to the leaves of young tomato plants.

Table 20

Effect of Calcium Chloride and Magnesium Sulfate on the Reduction of Urea Injury to Young Tomato Plants Grown in the Greenhouse (Second Test)

Materials and Concentrations Used in Each Solution	Average Score of Urea Injury[a]
.2 M urea alone	2.0
.2 M urea + .02 M $CaCl_2$	2.0
.2 M urea + .06 M $CaCl_2$	2.0
.2 M urea + .18 M $CaCl_2$	2.0
.4 M urea alone	5.0
.4 M urea + .02 M $CaCl_2$	5.0
.4 M urea + .06 M $CaCl_2$	5.0
.4 M urea + .18 M $CaCl_2$	5.0
.8 M urea alone	9.0
.8 M urea + .02 M $CaCl_2$	9.0
.8 M urea + .06 M $CaCl_2$	9.0
.8 M urea + .18 M $CaCl_2$	9.0
.18 M $CaCl_2$ alone	1.0
.4 M urea + .02 M $MgSO_4$	5.0
.4 M urea + .18 M $MgSO_4$	4.0
.18 M $MgSO_4$ alone	0.0

[a] Based on a scale from 0, which indicates no injury, to 10, which indicates the plants were injured beyond recovery.

In the fourth test magnesium sulfate was again tested together with zinc sulfate and ferrous sulfate and three unbuffered urea solutions adjusted to pH 6, 7, and 8. As the amount of magnesium sulfate was increased in urea solutions of the same molarity, the injury to the foliage appeared to drop proportionately. These results are not given in table form.

A more thorough study with magnesium sulfate added in a wide range of concentrations to urea solutions was made in the fifth test. That magnesium sulfate can reduce injury caused by urea to the foliage of tomato is evident from the results given in Table 21. It is interesting to note that with each of the three urea concentrations used the amount of injury was reduced as the magnesium sulfate content of the solutions increased. At the end of one week the plants that had been sprayed with a 0.5 M urea solution were almost devoid

Table 21

The Effects of Magnesium Sulfate on the Reduction of Urea Injury to Young Tomato Plants Grown in the Greenhouse (Fifth Test)

Materials and Concentrations Used in Each Solution	Average Score of Urea Injury[a]
.3 M urea alone	6.0
.3 M urea + .15 M $MgSO_4$	2.0
.3 M urea + .30 M $MgSO_4$	1.0
.3 M urea + .45 M $MgSO_4$	0.5
.4 M urea alone	7.0
.4 M urea + .15 M $MgSO_4$	2.0
.4 M urea + .30 M $MgSO_4$	1.5
.4 M urea + .45 M $MgSO_4$	1.0
.4 M urea + .60 M $MgSO_4$	0.5
.5 M urea alone	9.0
.5 M urea + .15 M $MgSO_4$	5.0
.5 M urea + .30 M $MgSO_4$	3.0
.5 M urea + .60 M $MgSO_4$	1.5
.5 M urea + .90 M $MgSO_4$	0.5
.3 M $MgSO_4$ alone	0.0
.6 M $MgSO_4$ alone	0.0

[a] Based on a scale from 0, which indicates no injury, to 10, which indicates that plants were injured beyond recovery.

of functional leaves. A representative leaf showing the extent of injury can be seen in Plate 1. When 0.5 mole of magnesium sulfate was added to a liter of 0.5 M urea solution, the plants suffered only minor injury to the tips and margins of the younger leaves (Plate II).

Plate 1

A Representative Leaf from a Tomato Plant Sprayed with a .5 M Urea Solution. Note Distortion and Necrosis.

Plate II

A Representative Leaf From a Tomato Plant Sprayed
with a .5 M Urea + .5 M $MgSO_4$ Solution.
Note Minor Injuries at Tips
and Margins.

Experiment VI Absorption of urea

Emmert and Klinker suggested that the foliar injury caused by urea could be reduced by increasing the carbohydrate level of the leaf. They succeeded by adding sucrose to urea solutions, in increasing the carbohydrate content of the leaf as well as reducing foliar injury. Cook later showed that sucrose actually reduced the amount of urea absorbed.

In Experiment V it was shown that injury to the foliage of tomato plants was reduced by the addition of magnesium sulfate to the urea solution. In this study an attempt was made to learn how reduction of injury is brought about when magnesium sulfate is added to urea solutions. Its effect on the rate of absorption of urea was studied in two separate tests in which plants were treated with a known amount of urea from either urea solutions without magnesium sulfate or urea solutions containing magnesium sulfate. At various intervals of time the amount of urea found in the leaf tissues, as well as the amount found on the leaf surfaces, was measured.

Materials and methods

Tomato plants were grown in eight-inch pots in a greenhouse to a height of approximately sixteen inches. Urea was applied to the surface of the leaves by brushing it on with camel hairs attached directly to the delivery end of a two-milliliter Mohr pipette. In this way it was possible to measure the amount of urea applied more accurately than by spraying.

Treatments for the first test consisted of applications of: (1) 0.1 M urea alone; (2) 0.1 M urea plus 0.1 M magnesium sulfate; and (3) a control receiving no urea. All but the control plants received approximately 9.6 milligrams of urea. At intervals of three, six, and twelve hours after treatment all leaves were collected from each plant and washed in three portions of water to remove unabsorbed urea. The total volume of wash solution for each sample amounted to one liter. The wash solution was evaporated on a hot plate to a volume of 20 milliliters. Urea was determined in the concentrated solution by a method adapted to micro-scales (2).

The leaves were then quickly dried at 80° Centigrade in a forced-draft oven and ground in a Wiley mill to pass through an eighty-mesh screen. Urea was determined on the finely-ground leaf tissue by the same method employed for the concentrated wash solution. The control plants were used to correct for the presence of free ammonia in the determination of urea in leaf tissues of the tomato plant.

A second test was conducted with a few modifications. Twelve milligrams of urea were applied to each plant. The experiment was designed to use the following treatments: (1) 0.2 M urea alone; (2) 0.2 M urea 0.4 M magnesium sulfate; and (3) a control receiving no urea. All treatments were replicated three times and the results analyzed statistically. Urea Determinations were made by the same methods employed in the first test.

Results

Urea solutions with and without magnesium sulfate were applied to the leaves of young tomato plants to determine the effect of magnesium sulfate on the rate of absorption of urea. The results of the first test are given in Table 22.

Table 22

The Effect of Magnesium Sulfate on the Rate of Absorption of Urea Applied to the Leaves of Young Tomato Plants (First Test)

Time Interval	Treatments[a]	Per Cent Urea in Leaf Tissue	Per Cent Urea on Leaf Surfaces	Total Per Cent Urea Recovered
3 Hrs.	A	31.0	48.2	79.2
	B	7.5	74.6	82.1
6 Hrs.	A	41.4	38.8	80.2
	B	12.9	68.2	81.1
12 Hrs.	A	48.8	26.6	74.5
	B	25.5	48.2	73.7

[a] A = 0.1 M urea alone.
B = 0.1 M urea + 0.1 M $MgSO_4$.

The addition of magnesium sulfate resulted in a reduction in the amount of urea absorbed. Of the total amount of urea applied 48.2 per cent was found on the leaf surfaces three hours after application on the plant treated with 0.1 M urea solutions alone. In the same time interval the plant treated with 0.1 M urea plus 0.1 M magnesium sulfate solutions was found to contain 74.6 per cent of the urea on the leaf surfaces. Similar results were obtained when the leaf tissue of those plants were analyzed for total urea (Table 22). In all cases the amount of urea found in the leaf tissue was larger in those plants receiving no magnesium sulfate in the urea solution.

In the second test all treatments were replicated three times. Rate of absorption studies were made at intervals of three, six, twelve, twenty-four hours, and seven days. In Table 23 is given the average per cent urea found on the leaf surfaces of the tomato plants at five intervals of time after application. Significant differences were shown at the .01 level between treatments, time intervals, and their first order interaction. Proportionately less urea was found on the leaf surfaces as the length of the absorption period was increased. The results showed that the addition of magnesium sulfate reduced the intake of urea by the leaves. Plants treated with urea solutions containing magnesium sulfate were found to have more unabsorbed urea on the leaf surfaces than plants treated with urea solutions without magnesium sulfate.

In Table 24 are given the results of analysis for urea in the leaf tissues of the same tomato plants.

Table 23

The Effect of Magnesium Sulfate on the Absorption of Urea as Measured by the Amount Found on Tomato Leaf Surfaces at Varying Intervals of Time After Application (Second Test)

Time Intervals	Treatments		
	0.2 M Urea Alone[a]	0.2 M Urea + 0.4 M $MgSO_4$[a]	Mean
3 Hours	37.9	61.2	49.6
6 Hours	34.6	42.2	38.4
12 Hours	25.4	32.8	29.1
24 Hours	8.2	27.1	17.7
7 Days	3.9	4.5	4.2
Mean	22.0	33.6	

L.S.D. for Treatment Means .01 = 4.2%

L.S.D. for Time Interval Means .01 = 5.6%

[a] Average of 3 replicates in per cent of total urea applied.

Statistical significance was also shown at .01 level for treatments, time intervals, and their first order interaction. Less urea was found in the tissue of plants treated with urea solutions containing magnesium sulfate than in the tissue of plants treated with urea solutions containing no magnesium sulfate.

In Table 24 it can be seen that as much as 24.7 per cent of the total urea applied was found in the leaf tissues

Table 24

The Effect of Magnesium Sulfate on the Amount of Urea Found in Tomato Leaf Tissues at Varying Intervals of Time After Application (Second Test)

Time Intervals	Treatments		Mean
	0.2 M Urea Alone[a]	0.2 M Urea + 0.4 M $MgSO_4$[a]	
3 Hours	28.6	3.9	16.3
6 Hours	31.6	22.2	26.9
12 Hours	34.6	25.6	30.1
24 Hours	50.0	29.9	40.0
7 Days	24.7	10.6	17.7
Mean	33.9	18.4	

L.S.D. for Treatment Means .01 = 1.9%

L.S.D. for Time Interval Means .01 = 6.0%

---

[a] Average of 3 replicates in per cent of total urea applied.

seven days after application.

Experiment VII Urease activity

The method by which the tomato plant utilizes urea after it has been taken into the leaf is not definitely known. The enzyme, urease, if present in an active form to a measurable extent, can hydrolyze urea to ammonia and carbon dioxide readily. The ammonia thus released is available for the formation of amino acid and the more complex proteins.

Detection of urease in an active form in the tomato leaf would offer a partial explanation of some of the processes involved in the metabolism of urea. It might also serve to explain why urea causes severe foliar injuries under certain conditions. A high rate of absorption, which has been shown experimentally, coupled with high urease activity could release quantities of ammonia large enough to be toxic to the plant.

This study was undertaken primarily to determine if urease is present in an active form in the tomato leaf and to determine its relationship to foliar injury and metabolism.

Materials and methods

Urease activity was determined by the aeration method described by Sumner and Hand (21) with several modifications. To a liter of phosphate-buffered solution, twenty grams of gum arabic were added to prevent inactivation of the enzyme. Temperature for the reaction was raised from 20° Centigrade to 30° Centigrade and the time was increased from four minutes to one hour or more. A two per cent boric acid solution was used to trap the ammonia and the latter was titrated with .0210 N hydrochloric acid with methyl red-methylene blue as indicator.

The technique was tested quantitatively before it was used for this study. A commercial preparation of jack bean meal was used as a source of urease. The determinations of urease activity from jack bean which were run with an excess of urea were sufficiently close to warrant the use of the method in this study. A control was included with each set of samples that were analyzed. It served to detect inactivation of the enzyme in each test. If the check, to which was added a commercial urease preparation, did not produce results equivalent to the quantitative tests made earlier, all of the determinations were discarded.

A method of preparation of leaf tissues was desired which would permit release of the enzyme from the cells, allowing it to come in direct contact with urea. Two methods were studied. For the first, leaf samples from six-week-old tomato plants were collected and frozen immediately in water. After thawing, the leaf tissue was macerated thoroughly in a Waring blendor for five minutes. An aliquot of the suspension was taken for analysis.

The second method of preparation consisted of drying the leaf tissue in a forced-draft oven at 40° Centigrade.

Upon drying, the leaf tissue was ground in a Wiley mill to pass through an eighty-mesh screen. In this way the tissue was reduced to a rather finely divided material. A portion was weighed and placed in suspension with a phosphate-buffered solution containing a known amount of urea for determination of urease activity.

A limited number of tests were made with roots and dried seed of tomatoes treated in the same manner as the determinations with dried leaf tissue.

Results

Samples of young tomato leaf tissue were frozen and then macerated in a Waring blendor for five minutes. Urease activity was measured on aliquots from suspensions of the macerated tissue. A blank was run on the same leaf tissues without the addition of urea. This accounted for free ammonia in the plant tissue and reagents. The titer value for the blank was subtracted from the sample value to give the net titer. It was assumed that the net titer value represented ammonia converted from urea by the urease present in the tomato leaf. A unit of urease activity is defined here as the number of milligrams of urea converted in one hour by one gram dry weight of the material. An average of .62 units of urease activity was obtained from six tests with tissue prepared by freezing and maceration (Table 25).

Urease activity was measured in tomato leaf tissue

Table 25

Urease Activity of Tomato Leaves Prepared by Freezing before Macerating in a Waring Blendor

Fresh Weight of Sample[a]	Period of Digestion	Titer (.0210 N HCl)	Units of Urease Activity[b]
Sept. 8, 1951			
5 gm.	1 Hr.	1.92	0.67
5 gm.	1 Hr.	1.95	0.71
5 gm. (blank)	1 Hr.	1.45	-
Sept. 10, 1951			
5 gm.	1 Hr.	1.82	0.54
5 gm.	1 Hr.	1.90	0.71
5 gm. (blank)	1 Hr.	1.40	-
Sept. 12, 1951			
5 gm.	1 Hr.	1.78	0.51
5 gm.	1 Hr.	1.85	0.60
5 gm. (blank)	1 Hr.	1.42	-
Average			0.62

[a] Dry Matter - 9%.

[b] Number of milligrams urea converted in one hour by gram of dry weight of material. Blank subtracted from sample to obtain net titer.

prepared by drying and grinding. Six tests made on three dates gave an average of 1.02 units of urease activity (Table 26). The time in these tests was extended to four hours after preliminary tests showed that in shorter periods the differences between blanks and test samples were too small to measure.

A few tests were made with frozen and macerated leaf

Table 26

Urease Activity of Tomato Leaves Prepared by Drying and Grinding

Dry Weight of Sample	Period of Digestion	Titer (.0210 N HCl)	Units of Urease Activity[a]
Oct. 3, 1951			
100 mg.	4 Hrs.	1.45	0.95
100 mg.	4 Hrs.	1.52	1.06
100 mg. (blank)	4 Hrs.	0.85	-
Oct. 4, 1951			
100 mg.	4 Hrs.	1.50	.89
100 mg.	4 Hrs.	1.53	.94
100 mg. (blank)	4 Hrs.	0.94	-
Oct. 8, 1951			
100 mg.	4 Hrs.	1.57	1.21
100 mg.	4 Hrs.	1.50	1.09
100 mg. (blank)	4 Hrs.	0.81	-
Average			1.02

[a] Number of milligrams urea converted in one hour by one gram of dry weight material. Blank subtracted from sample to obtain net titer.

tissue in an unbuffered solution. Instead of using a phosphate-buffered solution, enough distilled water was added to allow the leaf tissue homogeneate to come in contact with the urea. No urease activity was dectected under these conditions.

The results from tests made on two dates for urease activity in tomato roots are given in Table 27. The average of .47 units of urease activity was the lowest obtained for

Table 27

Urease Activity of Tomato Roots Prepared by Drying and Grinding

Dry Weight of Sample	Period of Digestion	Titer (.0210 N HCl)	Units of Urease Activity[a]
Sept. 24, 1951			
100 mg.	6 Hrs.	1.21	.52
100 mg.	6 Hrs.	1.17	.45
100 mg. (blank)	6 Hrs.	0.72	-
Sept. 25, 1951			
100 mg.	12 Hrs.	1.65	.48
100 mg.	12 Hrs.	1.58	.44
100 mg. (blank)	12 Hrs.	0.75	-
Average			.47

[a] Number of milligrams urea converted in one hour by one gram of dry weight of material. Blank subtracted from sample to obtain net titer.

any part of the tomato plant.

Tomato seeds were tested for urease activity by the same method employed in the root tissue. An average of .64 units of activity was obtained for four tests made on two dates (Table 28).

It can be seen from these results that a slight but positive measure of urease activity was obtained. In many cases it was necessary to take relatively large samples and to lengthen the time considerably to obtain positive results.

Table 28

Urease Activity of Tomato Seed
Prepared by Drying
and Grinding

Dry Weight of Sample	Period of Digestion	Titer (.0210 N HCl)	Units of Urease Activity[a]
Sept. 27, 1951			
200 mg.	6 Hrs.	2.18	.64
200 mg.	6 Hrs.	2.07	.59
200 mg. (blank)	6 Hrs.	.97	-
Sept. 28, 1951			
200 mg.	12 Hrs.	3.43	.64
200 mg.	12 Hrs.	3.52	.67
200 mg. (blank)	12 Hrs.	1.00	-
Average			.64

[a] Number of milligrams of urea converted in one hour by one gram dry weight of material. Blank subtracted from sample to obtain net titer.

DISCUSSION OF RESULTS

The experimental data were divided into two parts for presentation. The first part included studies to determine the response of tomatoes to urea sprays. The second part consisted of studies of interrelated problems on foliar injury, absorption, and urease activity. The results of all experiments in each part are discussed under those two headings.

Response of Tomatoes to Urea Sprays

The total nitrogen level of the leaf tissue is often considered as a good indication of the vigor of the plant. Results of studies under field and greenhouse conditions showed that it is possible to maintain a high level of leaf nitrogen in the tomato plant by foliar applications of urea. The maintenance of a satisfactory level of nitrogen in the leaf by foliar applications of urea is conditioned by the leaf area of the plant at the time of application of urea. This was demonstrated in comparative studies of the response of tomatoes to foliar and soil applications of nitrogen. The average increase in height of tomato plants receiving foliar sprays of urea was less during the first few weeks of growth than that from plants receiving equal amounts of nitrogen applied to the soil before planting. Much of the soil nitrogen was probably leached by heavy rains shortly after emergence of the plants above the surface of the soil. Yet, there appeared in the plots receiving soil applications to be sufficient nitrogen remaining in the soil to promote better growth during the next three weeks than in those receiving two foliar applications of urea.

Other measurements of the response of tomatoes to foliar sprays of urea indicate that the plant can absorb and utilize urea more effectively in the advanced stages of growth. The response, however, was not any more apparent than that from plants in plots receiving all of their nitrogen from soil applications. It must be pointed out here that rainfall conditions in the fall of 1950 were conducive to heavy leaching of soil nitrogen. Comparisons of weight and number of immature fruits showed that tomato plants were not more responsive to urea sprays than to similar amounts of nitrogen applied to the soil at the time of planting.

These results were corroborated by tests in the spring of 1951 and of 1952, comparing urea sprays with side-dressings of sodium nitrate as supplemental sources of nitrogen for tomatoes. The response, as measured by dates of maturity, weights, and numbers of fruits harvested, was not significantly different between the two sources of supplemental nitrogen. Rainfall during the 1951 growing season was limited, but was very heavy during the 1952 growing season. Under adverse weather conditions prevailing during the 1952 period, tomato plants did not respond more quickly to urea sprays than to sodium nitrate side-dressings.

The responses heretofore discussed were measured under field conditions where much of the material probably reached the soil to be absorbed by the roots. What part of the response attributed to spray treatment resulted from direct foliar absorption? Tests under controlled conditions in a greenhouse showed that foliar applications of urea increased height, fresh weight, and the total nitrogen level of the plants as compared with plants receiving no additional nitrogen. These results would indicate favorably that the tomato plant is able to absorb and utilize urea. Although the increases were statistically significant, the results were not too apparent to the observer. Even though the foliage of urea-sprayed plants was slightly darker green in color, the vigor that one would expect from six weekly applications of nitrogen was lacking.

Foliar Injury, Absorption, and Urease Activity

The resulting injury from foliar sprays of urea is quickly manifested in the tomato plant. It can be detected by the presence of dark, water-soaked areas on the leaves within three to four hours after application of urea sprays.

On the basis of reports that injury was reduced by the addition of sucrose, hydrated lime, and Bordeaux mixture, a number of other materials were tested. Magnesium sulfate, when added to urea solutions, was found to reduce injury to tomato plants.

Emmert and Klinker found that sucrose increased the carbohydrate level of the tomato plant. They suggested that the carbohydrates of the leaf combine with the urea to reduce foliar injury. They made no studies on the effect of sucrose on the rate of absorption of urea. Cook investigated this point and found that sucrose reduced the absolute amount of urea absorbed by the leaves in a definite period of time.

The results of studies on the effect of magnesium sulfate on the absorption of urea are in accord with those reported by Cook. In Tables 22, 23, and 24 it can be seen that magnesium sulfate reduced the rate of absorption of urea by tomato plants. These results were checked by determining the amount of urea on the leaf surfaces as well as in the leaf tissues at various intervals of time after application. It must be pointed out here that all of the urea which was applied to the plants was not recovered. Some of the urea could have been translocated to other parts of the plant or hydrolyzed either in the leaf tissue by the cells or on the leaf surfaces by microorganisms. Notwithstanding the inability to recover all of the urea, the results are considered sufficiently accurate to give some information on the questions at hand. Four to eight per cent only of the total urea applied was found three hours after application in the leaf tissues of those plants treated with urea plus magnesium sulfate solutions. In the same period of time, 28 to 31 per cent of the total urea applied was found in the leaf tissues of plants treated with urea solutions containing no magnesium sulfate.

It would appear from these results that the degree of foliar injury sustained by a tomato plant is directly proportional to the amount of urea absorbed within the first few hours after application. By reducing the rate of absorption of urea, magnesium sulfate serves to lessen foliar injury.

The mechanism by which magnesium sulfate reduces the rate of absorption of urea is not known. It is possible that it acts to reduce the permeability of the cell membranes to urea. It has been shown that some elements are capable of preventing or reducing the penetration of dyes into the cell. Brooks (4) found that potassium, calcium, sodium, and magnesium chlorides all checked the penetration of the dye dahlia into the living cells of Nitella.

Bayliss (3) clearly demonstrated the antagonistic effect of mixtures of salts upon the permeability of membranes. When a freshly-cut slice of beet was placed in 0.31 M solution of sodium chloride, the red pigment diffused out into the solution. This did not occur when a slice of beet was placed in water. This showed that sodium chloride increased the permeability of the membrane sufficiently to allow the red pigment of the cell to diffuse out. The outward diffusion of the red pigment was not detected when a slice of beet was placed in a 0.31 M sodium chloride plus 0.17 M calcium chloride solution.

Urease activity in different parts of the tomato plant was measured in the laboratory. No urease activity was found when macerated tomato leaf tissue was added to an unbuffered urea solution even though glass-distilled water and gum arabic were used. It was also measured in a phosphate-buffered solution at a pH of 7.0. Under these conditions a slightly positive measure for urease activity in different parts of the plant was obtained. The results from the two methods of preparation of leaf tissue showed that a higher value was obtained for urease activity when the leaf tissue was ground and dried as compared with freezing and maceration of the fresh leaves.

The presence of urease in the leaf tissues of the tomato plant was established when the reaction was carried on in a phosphate buffered solution. This does not prove that urease is active in the tissue of a growing plant. Two observations made in this study seem to indicate that there is little, if any, urease activity in the leaves of a growing tomato plant. First, no urease activity was noted when it was measured in an unbuffered system with frozen and macerated fresh tissue. Second, as much as 24.7 per cent of the total urea applied to the plants was found in the leaf tissues seven days after application. Urease, being an enzyme which is capable of hydrolyzing large quantities of urea in a certain period of time, probably would have hydrolyzed the few milligrams of absorbed urea in a much shorter period of time than seven days. It would appear from these results that sufficient ammonia was not released through enzymatic hydrolysis to cause foliar injury.

SUMMARY AND CONCLUSIONS

Nitrogen fertilization of tomatoes with foliar sprays of urea was evaluated from four tests under field and greenhouse conditions during 1950, 1951, and 1952.

Records taken during the three weeks following spray applications of urea showed that the average height increases of plants receiving urea sprays were significantly higher at the .01 level when compared with plants receiving no nitrogen. These increases, however, were not so large as those from plants in plots receiving all of their nitrogen from applications to the soil before planting. These results would indicate that foliar applications of urea to the tomato plant in the early stages of growth are less beneficial than equal amounts of nitrogen applied to soil.

As measured by weight and number of fruits produced, the response of tomatoes to foliar sprays of urea was comparable to equal amounts of nitrogen applied to the soil.

The application of foliar sprays of urea did not significantly affect date of maturity when compared with equal applications of nitrogen from side-dressings of sodium nitrate. Weight and number of tomatoes harvested did not differ between the two types of supplemental nitrogen.

When foliar sprays of urea were prevented from reaching the soil, the results showed that increases in height, fresh weight, and total nitrogen of plants receiving urea sprays were significantly higher at the .01 level than those of the control plants receiving no nitrogen. These results would indicate that the tomato plant is able to absorb urea and to utilize it for growth to a certain extent.

Heavy concentrations of urea caused severe injury to the foliage of tomatoes. Rate of absorption studies showed that urea was absorbed readily by the leaves. The degree of foliar injury sustained by the plant was proportional to the amount of urea absorbed by the leaves within three to four hours after application.

Magnesium sulfate added to the solution reduced foliar injury resulting from applications of strong urea solutions to tomato plants.

The rate of absorption of urea was reduced by the addition of magnesium sulfate to the spray solution. The mechanism by which the reduction is effected is not known.

One-fourth of the urea applied was found in the leaf tissue of tomato plants seven days after application.

Active urease was not detected in fresh leaf tissue homogeneates in an unbuffered system. In a phosphate-buffer, fresh leaf-tissue homogeneates gave a slightly positive measure for urease activity.

LITERATURE CITED

1. Association of Official Agricultural Chemists. Official and Tentative Methods of Analysis. 6th Ed. Washington, D. C. 1945.

2. Association of Official Agricultural Chemists. Official and Tentative Methods of Analysis. 7th Ed. Washington, D. C. 1950.

3. Bayliss, W. M. Principles of General Physiology. 4th Ed. Longmans, Green and Co. London. 1924.

4. Brooks, M. M. Studies on the permeability of living cells. VIII. The effect of chlorides upon the penetration of dahlia into *Nitella*. Protoplasma 2: 420-427. (Biol. abst. 2: 9384. 1948).

5. Cook, J. A. A study of some factors affecting the absorption of urea by McIntosh apple leaves. Unpublished Ph. D. dissertation, Cornell University. 1951.

6. Emmert, E. M. and J. E. Klinker. Spraying tomato foliage with sucrose to increase carbohydrates and protect against injury by urea sprays. Ky. Ag. Exp. Sta. Bul. 550. May, 1950.

7. Fisher, E. G., D. Boynton and K. Skodvin. Nitrogen fertilization of the McIntosh apple with leaf sprays of urea. I. Proc. Amer. Soc. Hort. Sci. 51: 23-32. 1948.

8. Fisher, E. G. and J. A. Cook. Nitrogen fertilization of the McIntosh apple with leaf sprays of urea. II. Proc. Amer. Soc. Hort. Sci. 55: 35-40. 1950.

9. Fleming, H. K. and R. B. Alderfer. The effects of urea and oil-wax emulsion sprays on the performance of the Concord grape vine under cultivation and in Ladino clover. Proc. Amer. Soc. Hort. Sci. 54: 171-176. 1949.

10. Granick, S. Urease distribution in *Soja max*. Plant Physiology 13: 29-54. 1938.

11. Franick, S. Urease distribution in *Canavalia ensiformis*. Plant Physiology 12: 601-623. 1937.

12. Haas, A. R. C. Experimental application of urea to lemon leaves. The Calif. Citrograph 34: 286, 318. 1949.

13. Hamilton, J. M., D. H. Palmiter and L. C. Anderson. Preliminary tests with Uramon in foliage sprays as a means of regulating the nitrogen supply of apple trees. Proc. Amer. Soc. Hort. Sci. 42: 123-126. 1943.

14. Harley, C. P., H. H. Moon and L. O. Regeimbal. Urea sprays in nitrogen nutrition of apple trees (preliminary report). Proceedings Cumberland-Shenandoah Fruit Workers Conference. 1948.

15. Harper, H. J. The accurate determination of nitrates in soils. Jour. Ind. Eng. Chem. 16: 180-183. 1924.

16. Jones, W. W. and E. R. Parker. Application of urea to foliage of orange trees. Calif. Citrograph 34: 463. 1949.

17. Mack, G. L. and N. J. Shaulis. Nutritional sprays on grapes. An abstract. Phytopath 37: 14. 1947.

18. Proceedings of the Twenty-Seventh Annual Meeting of the National Joint Committee on Fertilizer Application. Pages 37-51. 1951.

19. Proebsting, E. L. Nitrogen sprays: tests reported with fertilizer containing 44% organic nitrogen. Calif. Agric. 5 (3): 12. 1951 (Biol. abst. 25: 34721. 1951).

20. Reifer, I. and J. Melville. The source of ammonia in plant tissue extracts. II. The influence of urea. Jour. Biol. Chem. 178 (2): 715-726. 1949.

21. Sumner, J. B. and D. B. Hand. Determination of urease activity by aeration. Jour. Biol. Chem. 76: 149. 1928.

22. Sumner, J. B. and F. G. Somers. Chemistry and Methods of Enzymes. 2nd Ed. Academic Press Inc. New York, N. Y. 1947.

23. Weinberger, J. H., V. E. Prince and L. Havis. Tests on foliar fertilization of peach trees with urea. Proc. Amer. Soc. Hort. Sci. 53: 26-28. 1949.

24. Wentzler, J. E. and D. G. White. The effects of nitrogenous fungicides and insecticides on the chlorophyll content of apple leaves. Proc. Amer. Soc. Hort. Sci. 54: 81-85. 1949.

25. Yamaguchi, S. Studies on the resorption of urea by roots of Zea mays seedlings in sterile culture. Jour. Fac. Sci. Hakkaido Imp. Univ. Ser. 5. Bot. 1(1): 37-55. 1930. (From Biol. abs. 6: 24602).

APPENDIX

Table 1a

Fall, 1950 (Factorial) Analysis of Variance: Height of Plants. Two Methods of Nitrogen Application, 3 Levels Each, 4 Replications

Sources of Variance	Degrees of Freedom	Variance	F Values
Blocks	3	3.70	2.24
Soil Applications	2	104.67	63.44**
Spray Applications	2	11.45	6.94**
Soil x Spray	4	5.74	3.48*
Error	24	1.65	

Table 1b

Fall, 1950 (Factorial) Analysis of Variance: Average Total Nitrogen. Two Methods of Nitrogen Application, 3 Levels Each, 4 Replications, Leaves Sampled 4 Times

Sources of Variance	Degrees of Freedom	Variance	F Values
Blocks	3	0.0951	1.13
Soil Applications	2	8.4820	100.74**
Spray Applications	2	7.3210	86.95**
Soil x Spray	4	0.4020	4.77**
Error (a)	24	0.0842	
Dates	3	28.1453	411.48**
Dates x Soil	6	1.9338	55.55**
Dates x Spray	6	3.8000	28.27**
Dates x Soil x Spray	12	0.1221	1.79
Error (b)	81	0.0684	

Table 1c

Fall, 1950 (Factorial) Analysis of Variance: Weight of Immature Fruit. Two Methods of Nitrogen Application, 3 Levels Each, 4 Replications

Sources of Variance	Degrees of Freedom	Variance	F Values
Blocks	3	472,939	1.52
Soil Applications	2	4,227,737	13.55**
Spray Applications	2	1,308,574	4.19*
Soil x Spray	4	763,331	2.45
Error	24	312,015	

Table 1d

Fall, 1950 (Factorial) Analysis of Variance: Number of Fruits Set. Two Methods of Nitrogen Application, 3 Levels Each, 4 Replications

Sources of Variance	Degrees of Freedom	Variance	F Values
Blocks	3	125	1.16
Soil Applications	2	824	7.49**
Spray Applications	2	804	7.31**
Soil x Spray	4	294	2.67
Error	24	110	

Table le

Fall, 1950 (Factorial) Analysis of Variance: Air Dry Weight of Plant Tissue. Two Methods of Nitrogen Application, 3 Levels Each, 4 Replications

Sources of Variance	Degrees of Freedom	Variance	F Values
Blocks	3	42,507	1.03
Soil Applications	2	296,519	7.15**
Spray Applications	2	1,041,376	26.12**
Soil x Spray	4	31,085	-
Error	24	41,454	-

Table 2a

Spring, 1951 (Factorial) Analysis of Variance: Total Weight of Fruits. Four Split-applications of Nitrogen, 8 Replications

Sources of Variance	Degrees of Freedom	Variance	F Values
Blocks	7	158.8	1.15
Soil N Levels	1	309.2	2.23
Supplemental N Sources	1	314.4	2.27
Soil N x Supplemental N	1	93.0	-
Error	21	138.6	-

Table 2b

Spring, 1951 (Factorial) Analysis of Variance: Total Number of Fruits. Four Split-applications of Nitrogen, 8 Replications

Sources of Variance	Degrees of Freedom	Variance	F Values
Block	7	1,906	1.69
Soil N Levels	1	810	-
Supplemental N Sources	1	3,719	3.04
Soil N x Supplemental N	1	1,339	1.09
Error	21	1,225	-

Table 2c

Spring, 1951 (Factorial) Analysis of Variance: Effect of Split-applications of Nitrogen on Weight of Fruits Harvested in Early Period

Sources of Variance	Degrees of Freedom	Variance	F Values
Blocks	7	39.6	-
Soil N Levels	1	33.4	-
Supplemental N Sources	1	50.2	-
Soil N x Supplemental N	1	187.7	3.21
Error	21	58.5	-

Table 2d

Spring, 1951 (Factorial) Analysis of Variance: Effect of Split-applications of Nitrogen on Weight of Fruits Harvested in Late Period

Sources of Variance	Degrees of Freedom	Variance	F Values
Blocks	7	113.8	2.83
Soil N Levels	1	139.9	3.48
Supplemental N Sources	1	113.7	2.83
Soil N x Supplemental N	1	16.3	-
Error	21	40.2	-

Table 2e

Spring, 1951 (Factorial) Analysis of Variance: Average Total Nitrogen (per cent dry weight) of Tomato Plants. Four Split-applications of Nitrogen, 2 Dates of Sampling, 8 Replications

Sources of Variance	Degrees of Freedom	Variance	F Values
Blocks	7	0.08330	1.18
Soil N Levels	1	0.00600	-
Supplemental N Sources	1	0.51120	6.86*
Soil N x Supplemental N	1	0.00010	-
Error (a)	21	0.07450	-
Dates of Sampling	1	7.70070	246.58**
Dates x Soil N	1	0.05180	1.66
Dates x Supplemental N	1	0.34820	11.15**
Dates x Soil N x Supp. N	1	0.07710	2.47
Error (b)	28	0.03123	-

Table 3a

Spring, 1952 (Randomized-block) Analysis of Variance: Weight of Tomatoes from Four Supplemental Nitrogen Treatments. 4 Replications

Sources of Variance	Degrees of Freedom	Variance	F Values
Blocks	3	474.5	8.82**
Treatments	3	13.0	-
Error	9	53.8	-

Table 3b

Spring, 1952 (Randomized-block) Analysis of Variance: Number of Tomatoes Harvested from Four Supplemental Nitrogen Treatments. 4 Replications

Sources of Variance	Degrees of Freedom	Variance	F Values
Blocks	3	5,281.4	5.68*
Treatments	3	46.4	-
Error	9	929.6	-

Table 4a

Greenhouse, 1951, (Split-plot) Analysis of Variance: Average Increase in Height of Tomato Plants, 3 Concentrations of Urea, 3 Dates of Sampling, 4 Replications

Sources of Variance	Degrees of Freedom	Variance	F Values
Blocks	3	18.51	4.91*
Urea Concentrations	2	78.33	20.78**
Error (a)	6	3.77	-
Dates	2	318.91	30.87**
Dates x Urea Concentrations	4	0.38	-
Error	18	10.33	-

Table 4b

Greenhouse, 1951 (Split-plot) Analysis of Variance: Average Fresh Weight of Tomato Plants, 3 Concentrations of Urea, 3 Dates of Sampling, 4 Replications

Sources of Variance	Degrees of Freedom	Variance	F Values
Blocks	3	355.80	5.55*
Urea Concentrations	2	1272.12	19.83**
Error (a)	6	64.14	-
Dates	2	1845.85	19.57**
Dates x Urea Concentrations	4	126.94	1.35
Error (b)	18	94.31	-

Table 4c

Greenhouse, 1951 (Split-plot) Analysis of Variance: Average Total Nitrogen of Tomato Leaf Tissues, 3 Concentrations of Urea, 3 Dates of Sampling, 4 Replications

Sources of Variance	Degrees of Freedom	Variance	F Values
Blocks	3	0.03133	1.65
Urea Concentrations	2	1.61775	85.06**
Error (a)	6	0.01902	-
Dates	2	0.41025	40.34**
Dates x Urea Concentrations	4	0.18083	17.78**
Error (b)	18	0.01017	-

Table 6a

Second Test (Split-Plot) Analysis of Variance: Effect of $MgSO_4$ on Rate of Absorption of Urea. Two Treatments, 5 Time Intervals, 3 Replications

Sources of Variance	Degrees of Freedom	Variance	F Values
Blocks	2	1.20	-
Time Intervals	4	1875.65	223.8^{**}
Error (a)	8	8.38	-
Treatments	1	1012.69	76.7^{**}
T x T I	4	128.75	9.8^{**}
Error (b)	10	13.20	-

Table 6b

Second Test (Split-Plot) Analysis of Variance: Effect of $MgSO_4$ on the Amount of Urea Found in Leaf Tissues. Two Treatments, 5 Time Intervals, 3 Replications

Sources of Variance	Degrees of Freedom	Variance	F Values
Blocks	2	14.95	1.5
Time Intervals	4	564.19	58.1^{**}
Error (a)	4	9.72	-
Treatments	1	1794.14	666.0^{**}
T x T I	4	70.54	26.2^{**}
Error (b)	10	2.70	-

BIOGRAPHICAL ITEMS

James Montelaro was born on March 3, 1921, in Melville, Louisiana. His family moved to Eunice, Louisiana, in 1927. Upon completion of his secondary education at the Mamou High School in 1938, he entered Southwestern Louisiana Institute, where he was awarded a B. S. degree in Agriculture in 1941.

In the fall of 1941, he entered Louisiana State University as a graduate student in Horticulture. He was called to active duty by the Navy in August, 1942.

After having served as an officer aboard a minesweeper in the Pacific theater of war, he was returned to inactive duty in 1947.

In 1949 he re-entered Louisiana State University and received a Master of Science degree in 1950. Since that time he has pursued his studies at the University of Florida.

This dissertation was prepared under the direction of the chairman of the candidate's supervisory committee and has been approved by all members of the committee. It was submitted to the Graduate Council and was approved as partial fulfilment of the requirements for the degree of Doctor of Philosophy.

August 16, 1952

L. E. Grinter
Dean

SUPERVISORY COMMITTEE:

Chesley B. Hall
Co-chairman

Co-chairman

R. A. Dennison

Luther C. Hammond

F W Stearns

C. V. Noble